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Synthesis and anticonvulsant activity of some potent 5,6-bis aryl 1,2,4-triazines

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Abstract: In the present research, a series of 5,6-bis aryl 1,2,4-triazines **5a**~**5f** were synthesized by condensation of various benzils **4a**~**4f** with aminoguanidine bicarbonate and were screened in vivo, for their anticonvulsant and neurotoxicity studies. Compounds **5a**, **5b** and **5d** were found to be potent molecules of this series, when compared with the reference drugs phenytoin sodium, diazepam and lamotrigine. The structures of these compounds were established by IR, ^{1}H NMR, ^{13}C NMR and mass spectroscopic data.

Key words: 5,6-Bis aryl 1,2,4-triazines, Synthesis, Anticonvulsant activity
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INTRODUCTION

Epilepsy, characterized by the periodic and unpredictable occurrence of seizures is the most prevalent neurological disorder, affecting 0.5% to 1% of the worldwide population (45~100 million) (Chang and Lowenstein, 2003; McNamara, 2001). Conventional antiepileptic drugs (AEDs) though widely used by approximately 25% of epileptics (Sander, 1993; Pastalos, 1999) fail to adequately control seizures and exhibit unfavourable side effects such as ataxia, hepatotoxicity, gingival hyperplasia and megaloblastic anemia (Duncan, 2002; Danny *et al*., 1992).

Phenytoin and carbamazepine prolong the inactive state of the voltage-dependent Na^{+} channels. A similar action was found for felbamate and topiramate. Topiramate also affects chloride currents and increases the number of channel openings induced by GABA (gama amino butyric acid) (Petroff *et al*., 1995). Contrary to this lamotrigine acts by prolonging inactivation of voltage-sensitive Na^{+} channels. Selectivity differences for the α subunits of Na^{+} channels might be responsible for the two different mechanisms (Taylor, 1996). The 1,2,4-triazines seem to act by inhibition of the Na^{+} channel in a frequency-dependent manner. As proposed previously, the pharmacophoric elements responsible for this mechanism are lipophilic aryl ring and hydrogen bonding to triazine moiety. The attachment of a second aryl ring to increase the van der Waal's bonding at the binding site, substitutions by diverse electron rich groups on aryl ring has proven to increase potency in the maximal electroshock seizure (MES) screen (Pandeya *et al*., 2000; Dimmock *et al*., 1995a). These structural contemplations gave impetus to synthesize some novel and potent 1,2,4-triazines, with phenyl having substitutions at C-5 and C-6 with halogen groups (electron withdrawing), which were considered to have sufficient diverse electronic, steric and hydrophobic characters.

CHEMISTRY

The synthesis of various 1,2,4-triazines was

accomplished as shown in Fig.1, by condensation of aldehydes **1** and **2** to yield compounds **3a**~**3f**, which structures were confirmed by disappearance of a peak between δ 9.6 to 9.9 for −CHO proton (Erickson, 1952). Compounds **3a**~**3f** on reacting with c-HNO_3 (concentrated HNO_3) yield corresponding benzils **4a**~**4f**, which were confirmed by disappearance of a singlet of −OH between δ 2.0 to 2.3 and CH proton between δ 6.0 to 6.2 in the ^{1}H NMR spectra (Sawyer and Copp, 1992). Substituted benzil derivatives **4a**~**4f** on condensation with aminoguanidine bicarbonate yield corresponding 5,6-bis aryl 1,2,4-triazines **5a**~**5f**. The structures of **5a**~**5f** were established by the appearance of a singlet between δ 4.0 to 4.3 for 2 protons of $-NH_2$ in the ^{1}H NMR spectra and disappearance of the peak at δ 195.2 (C=O) which was present in **4a**~**4f** (Richard *et al*., 1972). The spectral data of each compound was shown in the EXPERIMENTAL PROTOCOLS section and physical data was presented in Table 1.

PHARMACOLOGY

All the 5,6-bis aryl 1,2,4-triazines **5a**~**5f**, obtained from the reactional sequence were injected intraperitoneally into mice and evaluated for acute toxicity test, initial anticonvulsant screening and minimal motor impairment by the rotorod (neurotoxicity, NT) test, with three dose levels (30, 100 and 300 mg/kg) at two different time intervals.

The profile of anticonvulsant activity was established by MES pattern test and subcutaneous pentylene tetrazole (ScPTZ) seizure threshold test. All the compounds **5a**~**5f** were evaluated for their CNS (central nervous system) behavioral activity in mice using actophotometer and Porsolt's swim pool test with rats.

RESULTS AND DISCUSSION

Compounds **5a**~**5f** were first tested for prelimi-

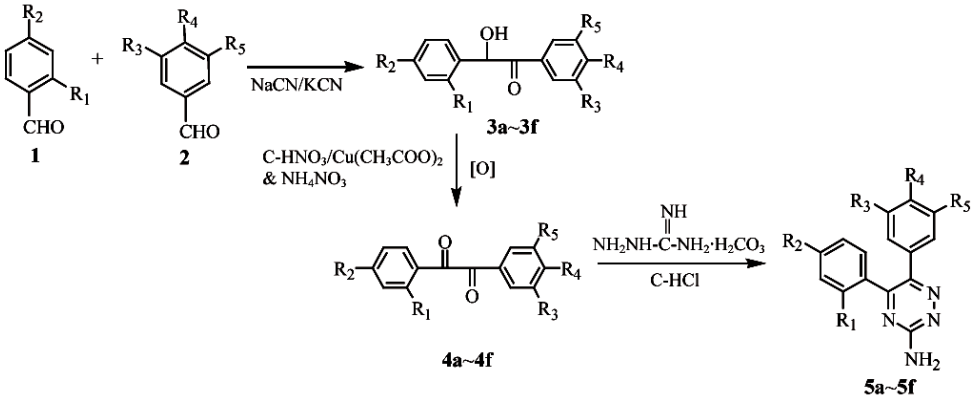

Fig.1 Synthesis of various 1,2,4-triazines

Table 1 Physical data and substitutions of synthesized 5,6-bis aryl 1,2,4-triazines

Compounds	MF	Elemental analysis (%)#			Substitutions					MW	Yield (%)	m.p. (°C)	CLOGP*
		C	H	N	R_1	R_2	R_3	R_4	R_5				
5a	$C_{17}H_{16}N_4$	73.89 (73.18)	5.84 (5.14)	20.27 (20.16)	H	CH_3	H	CH_3	H	276.3357	80	132~134	3.5864
5b	$C_{17}H_{17}N_5$	70.08 (70.11)	5.88 (5.16)	24.04 (24.23)	H	$N(CH_3)_2$	H	H	H	291.3504	78	94~96	3.0816
5c	$C_{18}H_{19}N_5O$	67.27 (67.17)	5.96 (5.91)	21.79 (21.11)	H	OCH_3	H	$N(CH_3)_2$	H	321.3764	85	110~112	2.9987
5d	$C_{16}H_{13}ClN_4O$	61.26 (61.14)	4.19 (4.18)	17.91 (17.18)	Cl	H	H	OCH_3	H	312.7536	74	198~200	3.9706
5e	$C_{17}H_{15}ClN_4O_2$	59.57 (59.47)	4.41 (4.14)	16.34 (16.27)	Cl	H	OCH_3	OCH_3	H	342.7796	70	212~214	3.0925
5f	$C_{18}H_{17}ClN_4O_3$	57.99 (57.23)	4.60 (4.16)	15.03 (15.34)	Cl	H	OCH_3	OCH_3	OCH_3	372.8056	71	172~174	2.8105

*Partition coefficient values are calculated by biobyte interactive log*P* calculator; #Data are express as calculated (found); MF: Molecular formula; MW: Molecular weight

nary anticonvulsant evaluation (Table 2) as per testing procedures described in National Institute of Neurological Disorders, Stroke, NH, Bethesda, MD, USA, for Anticonvulsant Screening Project (ASP). Compounds rendering protection in the MES test may prove to be useful in treating generalized tonic-clonic and complex partial seizures, and activity in the ScPTZ screening is deemed to denote the agents of value in treating seizures. The data in Table 3 reveal that all the compounds were active in both MES and ScPTZ screening at 30 min, thus indicating that synthesized compounds are rapid acting anticonvulsants. Neurotoxicity in mice was measured by the rotorod apparatus, with compounds **5d** and **5f** exhibiting neurotoxicity at dose of 300 mg/kg, when compared to other compounds.

In MES screening, all the compounds showed protection varying in the range of 8% to 84%, among them compounds **5a**, **5b**, and **5d** showed excellent protection. The characteristic feature of these compounds is substitution of electron rich atom/group at different positions of aryl ring, which demonstrates potent anticonvulsant activity in comparison to other derivatives (Dimmock *et al*., 1995b).

The bioevaluation data of the synthesized compounds by ScPTZ induced seizures led to comprehending why compounds **5b** and **5e** were equipotent in protection to phenytoin. The assessment led to understanding the importance of methoxy substitution on aryl ring, i.e. with dimethoxy, the optimum electron donor guaranteed superior anticonvulsant activity in **5e**. The introduction of 3rd methoxy group led to **5f**, which was found to be inactive. This observation suggests that the additional methoxy group caused interference with alignment at binding site. The optimum log*P* values in both **5b** and **5e** ensure lipophilicity required by the compounds. Although the drug levels in cerebrospinal fluid (CSF) or in

Table 2 Data of acute toxicity, initial anticonvulsant activity and minimal motor impairment test of the compounds (5a~5f) after intraperitoneal injection

Compound	Acute toxicity, ALD_{50} (mg/kg p.o.)	Intraperitoneal injection in mice[a] (mg/kg)					
		MES screen		ScPTZ screen		NT screen	
		0.5 h	4 h	0.5 h	4 h	0.5 h	4 h
5a	>1000	−	−	−	−	−	−
5b	>1000	−	−	−	−	−	−
5c	>1000	−	−	−	−	−	−
5d	>1000	−	−	−	300	−	300
5e	>1000	−	−	−	−	−	−
5f	>1000	−	−	−	−	−	300
Phenytoin	>1000	−	−	−	−	−	−

[a]The figures are the doses affording protection or causing neurotoxicity in 50% or more of the animals. The animals were examined 0.5 and 4 h after injections were made. The designation "−" indicates the absence of activity or toxicity at the maximum dose administered (300 mg/kg)

Table 3 Anticonvulsant activity data of compounds 5a~5f

Drug/test compounds	Dose	MES model[a]		ScPTZ model[a]		
		Duration of tonic hind limb extensor phase (s)	Animals protected (%)	Latency (s) (mean±*SEM*)	Duration of seizure (s) (mean±*SEM*)	Animals protected (%)
Control[b]	0.001 ml/g	13.43±0.32	0	135.8±2.18	248.19±2.56	0
5a	10	2.85±0.16	80	316.5±12.18	98.00±1.50	60
5b	10	4.56±0.19	76	3600	0	100
5c	10	3.25±0.38	67	234.67±5.61	126.89±1.29	49
5d	10	2.12±0.24	84	216.16±18.19	29.19±2.87	88
5e	10	9.48±0.36	30	3600	0	100
5f	10	12.46±0.13	8	373.7±13.39	126.33±0.28	50
Phenytoin[c]	40	0	100	−	−	−
Diazepam[d]	−	−	−	3600	0	100

N=6 in each group; [a]The compounds **5a**~**5f** were tested at a dose of 10 mg/kg; [b]0.5% methyl cellulose/water standard for control group; [c]Standard drug for MES pattern test; [d]Standard drug for ScPTZ seizure test

brain were not measured, higher log*P* values for the potent molecules suggest that there is a correlation between anticonvulsant activity and lipophilicity.

In the locomotor study using actophotometer (Table 4), the compound **5a** produced no behavioral despair effect when compared to phenytoin. Compound **5f** exhibited decreased locomotor activity in the 30 min interval, but did not exhibit significant behavior despair in 1 h time period. All other compounds were found to decrease locomotor activity. In a similar study using swim pool test, the immobility time after administration of the test compounds was compared with carbamazepine. The compound **5d** produced no significant CNS depression compared with control at $P<0.004$. All other tested compounds were found to emerge as CNS depressants, as they increased the immobility time (Table 5).

Table 4 Behavioral study data of compounds 5a~5f using actophotometer

Compound[a]	Activity score[b]		
	Control (24 h prior)	Post-treatment	
		0.5 h after	1 h after
5a	345±21.13	303±12.76*	278±17.19*
5b	234±21.34	45±12.12	48±12.81
5c	412±31.62	67±17.89	89±3.67
5d	342±45.18	47±24.19	76±12.66
5e	212±22.07	89±12.11	112±45.18
5f	247±10.90	127±13.45	207±54.43
Phenytoin[c]	265±21.37	54±12.78	58±12.55

[a]The compounds were tested at a dose of 10 mg/kg (i.p.); [b]Each score represents the mean±*SEM* of six mice, significantly different from the control score at $P<0.0001$, *$P<0.008$; [c]Tested at 5 mg/kg

Table 5 CNS depressant study data of compounds 5a~5f in forced swim pool test

Compounds[a] /drugs	Immobility time[b]	
	Control (24 h prior)	Post treatment (60 min after)
Control	173.67±13.69	178.67±22.3 NS
5a	123.56±11.49	189.58±55.5
5b	178.38±12.23	219.89±11.8
5c	138.78±10.39	256.88±19.9 NS
5d	108.78±10.19	161.89±10.3
5e	89.90±12.39	128.10±10.0
5f	119.76±11.18	157.89±16.7
Carbamazepine[a]	148.58±11.12	219.77±18.0

[a]The compounds were tested at a dose of 10 mg/kg (i.p.); [b]Each value represents the mean±*SEM* of six rats, significantly different from the control at $P<0.004$ and NS denotes not significant at $P<0.004$ (Student's *t*-test)

EXPERIMENTAL PROTOCOLS

Chemistry

Melting points were determined in open capillary tubes with a Thomas Hoover melting point apparatus and were uncorrected. Infrared spectra were recorded in KBr on (Nicolet MX-1) FTIR instrument, ^{1}H NMR spectra (in $CDCl_3$) were recorded by Varian Gemini 200 (200 Hz) NMR spectrometer. Electron impact (EI) mass spectra were determined on AMD-604 mass spectrometer operating at 70 eV.

1. 2-Hydroxy-1,2-diphenylethanone derivatives **3a**~**3f**

The equimolar mixture of compounds **1** and **2** (0.47 mol), rectified spirit (65 ml) and sodium cyanide (0.1 mol) in 50 ml of water was refluxed for 2 h. Then the mixture was cooled in ice bath and filtered, the solid thus obtained was washed with water and recrystalized with hot ethanol to obtain compounds **3a**~**3f**.

2. Substituted benzil derivatives **4a**~**4f**

A mixture of **3a**~**3f** (0.014 mol) and c-HNO_3 (2 mol) were refluxed until the oxides of nitrogen ceased to evolve and poured into ice-cold water, the solid thus obtained was filtered off and washed with water and recrystalized with ethanol to yield compounds **4a**~**4f**.

3. 5,6-Diphenyl-1,2,4-triazin-3-amine derivatives **5a**~**5f**

The mixture of aminoguanidine bicarbonate (0.08 mol), **4a**~**4f** (0.035 mol) and 30.0 ml of rectified spirit was acidified to pH 3~4 with c-HCl and refluxed for 2 h. Mixture was made alkaline with aq. ammonia and the solid so obtained was further recrystalized with ethanol.

Spectal data

1. 5,6-Dip-tolyl-1,2,4-triazin-3-amine **5a**

FT-IR (KBr): 1620.5 cm^{-1} (C=C), 3305.0 cm^{-1} ($-NH_{str}$), 1528.2 cm^{-1} ($C=N_{bend}$), 1350.2 cm^{-1} ($-CN_{str}$). ^{1}H-NMR ($CDCl_3$): δ 4.01 (s, 2H, $-NH_2$), δ 7.36 (d, 4H, −Ar-H), δ 7.12 (d, 4H, −Ar-H), δ 2.35 (s, 6H, $-2CH_3$). ^{13}C-NMR ($CDCl_3$ 125 MHz): 24.3 ($2CH_3$), 129.6 (4CH), 127.4 (4CH), 138.4 (2C), 130.1 (2C), 156.0 (C), 162.0 (C), 150.1 (C). Electron impact mass spectrum (*m*/*z*): 277.22 M^+ (32%), 206.09 ($C_{16}H_{14}$) (base), 120.01 (C_9H_{11}) (34.5%), 103.6 (C_9H_{11}) (22.4%), 90.05 (C_7H_7) (18.5%).

2. 5-(4-(Dimethylamino)phenyl)-6-phenyl-1,2,4-triazin-3-amine **5b**

FT-IR (KBr): 1580.2 cm^{-1} (C=C), 3480.2 cm^{-1} ($-NH_{str}$), 1538.5 cm^{-1} ($C=N_{bend}$), 1348.2 cm^{-1} ($-CN_{str}$). ^{1}H-NMR ($CDCl_3$): δ 4.09 (s, 2H, $-NH_2$), δ 6.65 (d, 2H, −Ar-H), δ 2.85 (s, 6H, $-CH_3$), δ 7.32 (d, 2H, −Ar-H), δ 7.22 (d, 1H, −Ar-H), δ 7.30 (d, 1H, −Ar-H), δ 7.48 (d, 2H, −Ar-H). ^{13}C-NMR ($CDCl_3$ 125 MHz): 114.8 (2CH), 128.4 (2CH), 129.3 (2CH), 127.5 (2CH), 128.8 (CH), 40.3 ($2CH_3$), 162.0 (C), 156.9 (C), 133.1 (C), 122.6 (C), 156.0 (C). Electron impact mass spectrum (*m*/*z*): 291.22 M^+ (base) (100%), 221 ($C_{16}H_{15}N$) (22%), 205.26 ($C_{14}H_{17}N$) (24.5%).

3. 6-(4-(Dimethylamino)phenyl)-5-(4-methoxyphenyl)-1,2,4-triazin-3-amine **5c**

FT-IR (KBr): 1586.4 cm^{-1} (C=C), 3475.6 cm^{-1} ($-NH_{str}$), 1519.5 cm^{-1} ($C=N_{bend}$), 1317.6 cm^{-1} ($-CN_{str}$) 1260.5 cm^{-1} ($-Ar\text{-}OCH_{3str}$), 2845.5 cm^{-1} ($-CH\text{-}OCH_{3str}$). ^{1}H-NMR ($CDCl_3$): δ 4.02 (s, 2H, $-NH_2$), δ 7.37 (d, 2H, −Ar-H), δ 7.30 (d, 2H, −Ar-H), δ 6.65 (d, 2H, −Ar-H), δ 6.83 (d, 2H, −Ar-H), δ 3.73 (s, 3H, $-OCH_3$), δ 2.85 (s, 6H, $-CH_3$). ^{13}C-NMR ($CDCl_3$ 125 MHz): 40.9 ($2CH_3$), 55.9 (CH_3), 114.8 (4CH), 128.4 (4CH), 149.6 (C), 160.7 (C), 125.4 (C), 122.6 (C), 156.1 (C), 162.0 (C), 150.9 (C). Electron impact mass spectrum (*m*/*z*): 321.15 M^+ (98.7%), 277.13 ($C_{17}H_{15}N_3O$) (32%), 251.13 ($C_{16}H_{15}N_3$) (41.5%).

4. 5-(2-Chlorophenyl)-6-(4-methoxyphenyl)-1,2,4-triazin-3-amine **5d**

FT-IR (KBr): 1603.6 cm^{-1} (C=C), 3470.8 cm^{-1} ($-NH_{str}$), 1508.7 cm^{-1} ($C=N_{bend}$), 1317.6 cm^{-1} ($-CN_{str}$) 1264.3 cm^{-1} ($-Ar\text{-}OCH_{3str}$), 2861.3 cm^{-1} ($-CH\text{-}OCH_{3str}$), 1087.6 cm^{-1} ($-C\text{-}Cl_{str}$). ^{1}H-NMR ($CDCl_3$): δ 4.23 (s, 2H, $-NH_2$), δ 3.73 (s, 3H, $-OCH_3$), δ 7.37 (d, 3H, −Ar-H), δ 6.83 (d, 2H, −Ar-H), δ 7.26 (m, 2H, −Ar-H), δ 7.49 (s, H, −Ar-H). ^{13}C-NMR ($CDCl_3$ 125 MHz): 55.9 ($-OCH_3$), 128.9 (3CH), 114.8 (2CH), 125.6 (CH), 130.6 (CH), 12.4 (CH), 134.5 (2C), 160.7 (C), 125.4 (C), 150.1 (C), 162.1 (C), 156.0 (C). Electron impact mass spectrum (*m*/*z*): 312.07 M^+ (100%), 242.13 ($C_{15}H_{11}ClO$) (base) (69%), 200.21 ($C_{10}H_9N_4O$) (13.16%).

5. 5-(2-Chlorophenyl)-6-(3,4-dimethoxyphenyl)-1,2,4-triazin-3-amine **5e**

FT-IR (KBr): 1601.4 cm^{-1} (C=C), 3337.8 cm^{-1} ($-NH_{str}$), 1508.7 cm^{-1} ($C=N_{bend}$), 1326.6 cm^{-1} ($-CN_{str}$) 1276.3 cm^{-1} ($-Ar\text{-}OCH_{3str}$), 2968.4 cm^{-1} ($-CH\text{-}OCH_{3str}$), 1043.6 cm^{-1} ($-C\text{-}Cl_{str}$). ^{1}H-NMR ($CDCl_3$): δ 3.73 (s, 6H, $-2OCH_3$), δ 4.36 (s, 2H, $-NH_2$), δ 6.93 (d, 1H, −Ar-H), δ 6.88 (s, 1H, −Ar-H), δ 6.72 (d, 1H, −Ar-H), δ 7.49 (s, 1H, −Ar-H), δ 7.36 (d, 1H, −Ar-H), δ 7.23 (m, 2H, −Ar-H). ^{13}C-NMR ($CDCl_3$ 125 MHz): 56.2 ($-2OCH_3$), 112.3 (CH), 115.8 (CH), 120.8 (CH), 127.4 (CH), 128.9 (CH), 127.4 (CH), 128.9 (CH), 130.7 (CH), 125.6 (CH), 134.9 (2C), 150.9 (C), 149.8 (C). Electron impact mass spectrum (*m*/*z*): 342.08 M^+ ($C_{17}H_{15}ClN_4O_2$) (base) (100%), 307.13 ($C_{14}H_{14}N_4O_2Cl$) (69.2%), 272.13 ($C_{16}H_{13}ClO_2$) (86.5%).

6. 5-(2-Chlorophenyl)-6-(3,4,5-trimethoxyphenyl)-1,2,4-triazin-3-amine **5f**

FT-IR (KBr): 1580.4 cm^{-1} (C=C), 3447.3 cm^{-1} ($-NH_{str}$), 1510.8 cm^{-1} ($C=N_{bend}$), 1329.7 cm^{-1} ($-CN_{str}$) 1254.2 cm^{-1} ($-Ar\text{-}OCH_{3str}$), 2968.4 cm^{-1} ($-CH\text{-}OCH_{3str}$), 1043.6 cm^{-1} ($-C\text{-}Cl_{str}$). ^{1}H-NMR ($CDCl_3$): δ 3.73 (s, 9H, $-3OCH_3$), δ 4.66 (s, 2H, $-NH_2$), δ 6.44 (s, 2H, −Ar-H), δ 7.49 (s, 1H, −Ar-H), δ 7.36 (d, 1H, −Ar-H), δ 7.26 (m, 2H, −Ar-H). ^{13}C-NMR ($CDCl_3$ 125 MHz): 56.5 ($-2OCH_3$), 56.9 ($-OCH_3$), 104.6 (CH), 128.3 (CH), 137.4 (CH), 130.4 (CH), 126.3 (CH), 134.5 (C), 151.3 (C), 139.2 (C). Electron impact mass spectrum (*m*/*z*): 372.08 M^+ (32%), 337.28 ($C_{18}H_{17}N_4O_3$) (base) (92%), 302.13 ($C_{17}H_{15}ClO_3$) (19.2%), 287.32 ($C_{14}H_{11}ClN_4O$) (26.5%).

Pharmacology

1. Acute toxicity test

The compounds were investigated for their acute toxicity (ALD_{50}) in albino mice by following the method of Smith (1960).

2. Neurotoxicity screening (Grundmann and Kreutzberger, 1954; Hosford and Wang, 1997)

Minimal motor impairment was measured in mice by the rotorod apparatus. The mice were trained to stay on an accelerating rotorod rotating at 10 r/min with rod diameter being 3.2 cm. The trained animals were injected intraperitoneally with the test compounds **5a**~**5f** at doses 30, 100 and 300 mg/kg. Neurotoxicity was indicated by the inability of the animal to maintain equilibrilium on the rod for at least 1 min in each of the three trials.

3. Anticonvulsant activity

(1) MES test (Kubicki and Codding, 2001; Stephani, 1989)

This activity was performed according to the reported method on albino rats of either sex, weighing between 80 to 120 g. Rats were divided into 8 groups of 6 animals each. The rats were treated with compounds **5a**~**5f** at dose of 10 mg/kg or phenytoin 40 mg/kg i.p. After 1 h they were subjected to shock of 150 mA by convulsiometer through ear electrodes for 0.2 s, the duration of extensor phase response was noted and animals in which extensor response was abolished were taken as protected rats.

(2) ScPTZ induced seizures test (Fisher, 1989)

The test was performed using albino mice which were divided into eight groups of 6 each. Pentylenetetrazole (80 mg/kg) (convulsant), diazepam (standard drug) and test compounds **5a**~**5f** were dissolved in 0.5% methyl cellulose/water and injected intraperitoneally at volume of 0.01 ml/g.

The time needed for the development of unequivocal sustained clonic seizure activity involving limbs (isolated myoclonic jerks or other preconclusive chewing behavior were not counted) and duration of seizure was carefully noted. Seizure free period of 1 h was considered as protection. The number of animals protected in each group was recorded and percentage of protection was calculated.

4. Locomotor activity (Stephani, 1989)

The titlered compounds **5a**~**5f** at 10 mg/kg were screened for their behavioral effects using actophotometer at 30 min and 1 h after injection. The locomotor activity was recorded by photocell as a digital score, and increased scores suggest good behavioral activity. The control was administered 0.5% methyl cellulose/water in a volume of 0.01 ml/g.

5. CNS depressant study

The forced swim pool method described in Porsolt *et al*.(1978) was followed. Albino rats were placed in a chamber (diameter: 45 cm, height: 20 cm) containing water up to a height of 15 cm at (25±2) °C. Two swim sessions were conducted, an initial 15 min pre-test, followed by a 5 min test session 24 h later. The animals were treated with intraperitoneal injection of test compounds at dose of 10 mg/kg before 30 min of test session. The period of immobility (passive floating without struggling, making only those movements which are necessary to keep its head above the surface of water) during the 5 min test period was measured.

CONCLUSION

In summary, this study highlights the importance of the structural features responsible for the anticonvulsant activity; *p*-methoxy derivative **5e** was found to be most potent in both anticonvulsant activity-evaluating models, which serve as prototypic compounds for subsequent molecular modifications.

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References

Chang, B.S., Lowenstein, D.H., 2003. Mechanisms of disease: epilepsy. *N. Eng. J. Med.*, **349**(13):1257-1262. [doi:10.1056/NEJMra022308]

Danny, D.S., René, H.L., Jill, L.S., Alan, V., 1992. Comparative anticonvulsant potency and pharmacokinetics of (+)- and (−)-enantiomers of stiripentol. *Epilepsy Res.*, **1**(11): 29-36.

Dimmock, J.R., Sidhu, K.K., Tumber, D.F., 1995a. Some aryl semicarbazones possessing anticonvulsant activitie. *Eur. J. Med. Chem.*, **30**(4):287-301. [doi:10.1016/0223-5234(96)88237-7]

Dimmock, J.R., Vashishtha, S.C., Stables, J.P., 1995b. Evaluation of the semicarbazones, thiosemicarbazones and bis-carbohydrazones of some aryl alicycylic ketones for anticonvulsant and other biological propertie. *Eur. J. Med. Chem.*, **30**(4):303-314. [doi:10.1016/0223-5234(96)88238-9]

Duncan, J.S., 2002. The promise of new antiepileptic drugs. *Br. J. Clin. Pharmacol.*, **53**(2):123-131. [doi:10.1046/j.0306-5251.2001.01540.x]

Erickson, J.G., 1952. 3-Amino-as-triazines. *J. Am. Chem. Soc.*, **74**(18):4706. [doi:10.1021/ja01138a506]

Fisher, R.S., 1989. Animal models of the epilepsies. *Brain Research Reviews*, **14**(3):245-278. [doi:10.1016/0165-0173(89)90003-9]

Grundmann, C., Kreutzberger, A., 1954. 1,3,5-Triazine. *J. Am. Chem. Soc.*, **76**(2):632-633. [doi:10.1021/ja 01631a107]

Hosford, D.A., Wang, Y., 1997. Utility of the lethargic (*lh*/*lh*) mouse model of absence seizures in predicting the effects of lamotrigine, vigabatrin, tiagabine, gabapentin, and

topiramate against human absence seizures. *Epilepsia*, **38**(4):408-414. [doi:10.1111/j.1528-1157.1997.tb01729.x]

Kubicki, M., Codding, P.W., 2001. Hydrogen bonding patterns in 3,5-diamino-6-aryl triazines. *Journal of Molecular Structure*, **570**(1-3):53-60. [doi:10.1016/S0022-2860(01)00477-X]

McNamara, J.O., 2001. Drugs Effective in the Therapy of the Epliepsies. *In*: Hardman, L.G., Limbird, L.E. (Eds.), Goodman and Gilman's the Pharmacological Basis of Therapeutics. McGraw Hill, New York, p.521-547.

Pandeya, S.N., Yogeswari, P., Stables, J.P., 2000. Synthesis and anticonvulsant activity of 4-bromophenyl substituted aryl semicarbazones. *Eur. J. Med. Chem.*, **35**(10):879-886. [doi:10.1016/S0223-5234(00)01169-7]

Pastalos, P.N., 1999. Synthesis of some oxime ether derivatives of 1-(2-naphthyl)-2-(1,2,4-triazol-1-yl)ethanone and their anticonvulsant and antimicrobial activities. *Curr. Opin. CPNS Invest. Drugs*, **1**(64):549-555.

Petroff, O.A., Rothman, D.L., Behar, K.L., Mattson, R.H., 1995. Initial observations on effect of vigabatrin on in vivo ^{1}H spectroscopic measurements of γ-aminobutyric acid, glutamate, and glutamine in human brain. *Epilepsia*, **36**(5):457-464. [doi:10.1111/j.1528-1157.1995.tb00486.x]

Porsolt, R.D., Anton, G., Blavet, N., Jalfre, M., 1978. Behavioural despair in rats: a new model sensitive to antidepressant treatments. *Eur. J. Pharmacol.*, **47**(4):379-391. [doi:10.1016/0014-2999(78)90118-8]

Richard, W.A., Peter, B.R., Theodre, J.F., 1972. Antimalarial activities of some 3,5-diamino-as-triazine derivatives. *J. Med. Chem.*, **15**(4):859-871.

Sander, J.W.A.S., 1993. Some aspects of prognosis in the epilepsies: a review. *Epilepsia*, **34**(6):1007-1016. [doi:10.1111/j.1528-1157.1993.tb02126.x]

Sawyer, D.A., Copp, F.C., 1992. Synthesis of novel triazoles as potent anticonvulsants. *Chem. Abstr.*, **108**:1125.

Smith, Q.E., 1960. Pharmacological Screening Tests Progress in Medicinal Chemistry. 1: Butterworths. London, p.1-33.

Stephani, U., 1989. The natural history of myoclonic astatic epilepsy the natural history of myoclonic astatic epilepsy. *Epilepsia*, **47**(Suppl. 1):S53-S55.

Taylor, C.P., 1996. Voltage-gated Na^{+} channels as targets for anticonvulsant, analgesic and neuroprotective drugs. *Curr. Pharm. Des.*, **2**(4):375-379.